No. 7 September 1998

BARLEY YELLOW DWARF *Newsletter*

CIMMYT

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Editor: M. Henry

CIMMYT

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CIMMYT is an internationally funded, nonprofit scientific research and training organization. Headquartered in Mexico, the Center works with agricultural research institutions worldwide to improve the productivity and sustainability of maize and wheat systems for poor farmers in developing countries. It is one of 16 similar centers supported by the Consultative Group on International Agricultural Research (CGIAR). The CGIAR comprises over 50 partner countries, international and regional organizations, and private foundations. It is co-sponsored by the Food and Agriculture Organization (FAO) of the United Nations, the International Bank for Reconstruction and Development (World Bank), the United Nations Development Programme (UNDP), and the United Nations Environment Programme (UNEP).

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Responsibility for this publication rests solely with CIMMYT.

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Contents

Foreword

The last BYD Newsletter (BYD-NL6) was sent in July 1997 to all the contributors and to most scientists from developing countries listed in the 1994 mailing list. A questionnaire on the future of the Newsletter and a subscription form were also included. The Newsletter was sent to 172 scientists or institutions and the questionnaire to 210 people.

Publication of this newsletter was delayed by several months due to a few responses received by the deadline for subscription (31 Dec. 1997). By now, around 35 responses have been received, indicating a 15% reply rate. Continuation of the publication was favored in 95% of the cases. The majority of answers favored an annual publication, keeping the same format, with occasional publication of related articles in the areas of cereal aphid resistance, biology of cereal aphids and, possibly, other luteoviruses. Several scientists from the developing world also requested references on recent BYDV publications.

In light of this questionnaire, the next issue of the BYD Newsletter will be published in one year, keeping in mind its informal nature. Whenever possible, extra information will be given on anything useful to BYDV researchers, such as tips and techniques, meetings, conferences, name of BYD researchers, etc. An informal review of BYDV publications printed the previous year will be added, but with no intention of being exhaustive. The review will be based on what appears in Current Contents, the AGRIS database and any papers I come across.

My main concern about the future of the BYD Newsletter is the low number of written contributions submitted. As you no doubt noticed, BYD-NL7 is very thin. In 1998, only seven papers were received from scientists outside CIMMYT. This will greatly affect the quality of the present issue.

I still believe that the Newsletter is a useful form of communication among scientists, especially in the developing world. I shall be grateful to anyone who contributes material for the next issue of the Newsletter. This could be in the form of country reports, recently published results, tips and any other subject that would contribute to the body of BYDV research, with a special focus on the developing world. Don't hesitate to promote the Newsletter, if you think it is useful. With the advent of the Internet, access to literature has been facilitated; however, there are still numerous places in the world where access to such tool is not easy, many of them being areas targeted by CIMMYT. However, if there is no interest in the Newsletter, we will have to stop its publication.

Because of the small size of this issue, every subscriber to BYD-NL7 will receive the next issue free of charge. I thank you all for the quality of the contributions and the support you have given the Newsletter throughout the years. I hope that the Yellow Dwarf will have a long future.

- **Note on citing *Newsletter* articles**

The *BYD Newsletter* consists of informal reports presented to foster communication and the exchange of ideas and information between developing and industrialized world BYD researchers. Responsibility for the views expressed in the article rests with each author. Research information distributed through the *Newsletter* is in the form of "notes" and not "published" in the sense of a refereed journal. It should NOT be cited in other publications without the authors' consent. Citations in refereed publications, whenever permitted, refer to the *BYD Newsletter* notes in the *text*, rather than in the bibliography. For example, specify "B. Yellow, *BYD Newsletter* 7: 11, 1998; unpublished data", or cite as "personal communication" (with the colleague's consent).

- ***BYD Newsletter* No. 8, 1999**

Contributions to BYD-*Newsletter* No. 8 should be received by **M. Henry** no later than ***June 30, 1998.*** Please send maximum one page per abstract. Abstracts should be written as narrative reports, including the most important progress made. A maximum of one figure and/or 2 tables can be accepted per abstract, if essential. Please follow the abstract header format and the citation format (e. g.: [Yellow and Dwarf, *Gen. Res. Crop. Evol.* 43:31-34 (1996)] and [Luteovirus et al., *J. Gen. Virol.* 65:222-229 (1995)].

To facilitate the edition of the Newsletter, submission of abstracts via electronic mail (m.henry@cgnet.com) or on diskette (ASCII-, WordPerfect- or Word for Windows file) is strongly encouraged.

- **Subscribe to the *BYD Newsletter* electronically**

The BYD Newsletter is **available on the Internet** on the CIMMYT home page (http://www.cimmyt.mx/) or on CGIAR home page (http://www.cgiar.org). Subscription and submission of contributions can be done through the Internet.

The mailing list included in BYD-NL7 is short due to the low rate of reply to subscription. In order to update it, I need your participation. Many of you, might be interested in receiving only the electronic form of the Internet. No charge will be cover for this version. However, if you want your name to appear in the mailing list, PLEASE FILL AND RETURN THE SUBSCRIPTION FORM included in this issue or electronically. I see the mailing list as a tool to increase communication and collaboration between BYDV scientists. Furthermore, several subscribers have requested the list of BYDV researchers. However, to be more accurate, this list need to be updated by the scientists still working in the BYDV area.

- **Funding the *BYD Newsletter***

The subscription to the *Newsletter* has been maintained to $15 for the past two issues. The contributions received for NL6 just cover its costs of printing and mailing, considering that it was sent to a large numbers of scientists in the developing world who benefited from subscription relief. The 1997 issue (NL7), due to its mall size can also be cover by the subscription fees received. I suggest to drop the cost of the 1999 to $10 because costs will be now reduced through the use of Internet.

BYDV tolerance and multiple disease resistance in *Triticum durum* interspecific derivatives

A. Comeau[1], C. A. St-Pierre[2], B. Kol[2], S. Pouleur[1], and R. Paquet[1]

[1]Agriculture and Agri-Food Canada, Sainte-Foy, Quebec G1V 2J3, Canada; [2]Dept. Phytologie, Universite Laval, Sainte-Foy, Que. G1K 7P4, Canada

Over the last 20 years, *Triticum durum* has always been quoted as more sensitive to BYDV than bread wheat, *Triticum aestivum*. From 1981 to 1991 our own efforts aimed at developing BYDV resistant germplasm, met with very little success in durum wheat. The durum line 82 PCD-476 was chosen as the most reliable moderately tolerant check. It was selected after 10 years of trials but it was much inferior in tolerance to Maringa bread wheat or any barley possessing the Yd_2 gene.

In Quebec, the climate is often humid, and durum wheat develops a broad range of other disease problems in those conditions, including scab (Fusarium head blight), root rot and other fungal diseases.

Tolerance vs resistance. Recognizing that tolerance differs from resistance, we attempted in 1983-86 to see if resistance genes, selectable by ELISA, would allow faster progress. Alien sources contain high resistance (seen as low ELISA values) and also good tolerance to BYDV. Alien introgression was attempted as a way to improve tolerance and resistance of bread and durum wheat faster. The field trials on yield losses would often reveal that non-resistant wheat had quite good tolerance.

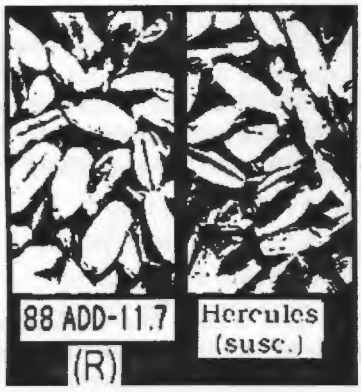

Fig.1. Seed quality comparison (visual) in a year of scab epidemic, 1996

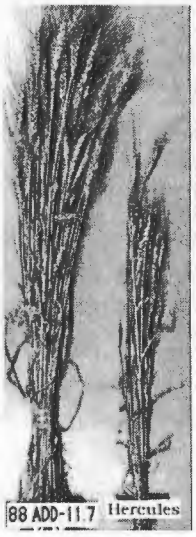

Fig. 2. Vigor of BYDV inoculated plants (1996)

One of the best tolerant lines was an interspecific-derived *Triticum aestivum* line from China, Long Miai 10, which is putatively derived from wheat x *Thinopyrum intermedium*. Interestingly enough, Long Miai 10 had ELISA levels generally higher than Maringa bread

wheat (let us note Maringa also has rather high ELISA values, and is used as BYDV tolerant check in our trials).

Introgression process. This line, Long Miai 10, was therefore used as a parent in crosses to both *T aestivum* and *T. durum*. The progenies were backcrossed to *T aestivum* and *T. durum*, selfed, and selected during the segregating generations F_3-F_5 using artificial BYDV inoculations. The first three cycles of selection of the *T. durum* trispecific populations showed very little promise. Out of about 8000 plants per year, not a single one seemed superior to 82 PCD-476 for BYDV tolerance. Then in the summer of 1993, about 10 plants that looked like "escapes" were harvested, from a cross labeled 88ADD-11. In this cross, line 82 PCD-476 is the recurrent durum parent. The good looking plants were multiplied for replicated trials.

In 1994 and 1995, the rows showed vigor and general disease resistance. In 1996, contrast was extreme between all the conventional durum wheat, which was severely affected by BYDV, and the 88ADD-11 re-selections which were as tolerant as Yd_2 barleys (Figs. 1, 2, 3). The 88ADD-11 series also showed better scab resistance than conventional *T. durum* in 1996. In 1997, the germination was uneven due to drought, but still the "88ADD-11" interspecific lines were the best BYDV tolerant (Fig. 4).

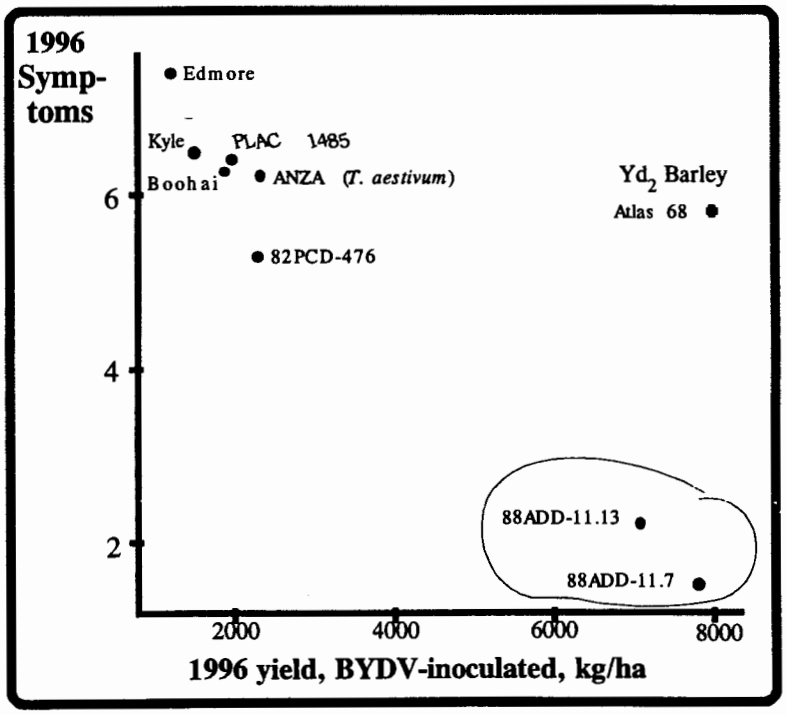

Fig. 3. Yield and symptoms of BYDV inoculated lines of *T. durum* in 1996, compared to *T. aestivum* Anza (mod. tol.), and Atlas 68 barley (resistant). The "88ADD-11" lines had low symptoms and high yield.

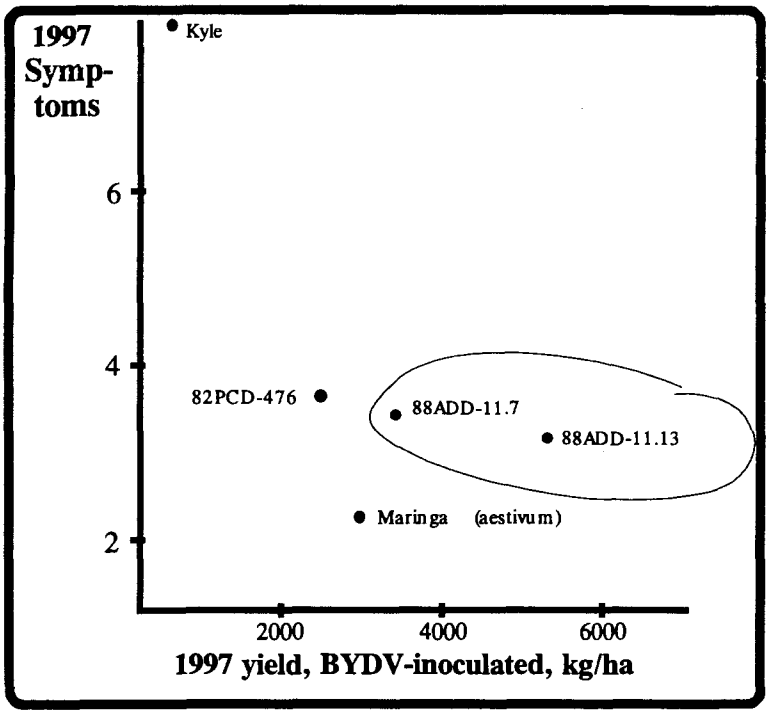

Fig. 4. Yield and symptoms of BYDV inoculated lines of *T. durum* in 1997, compared to *T. aestivum* Maringa (tolerant). The "88ADD-11" lines had low symptoms and high yield.

Observations on roots. The root system of the interspecific durum lines seemed more healthy than that of conventional durum, and visual inspection of roots revealed fewer root rot lesions. We are increasingly aware that one effect of artificial BYDV selection is to improve root health in many ways. But in this case, the root health effect seemed more important and desirable, as all conventional durum wheat lines become very sensitive to root rot by flowering time, in our growing area.

We also wondered if the root morphology might be different, and evaluated this hypothesis in tests done in long acrylic tubes, 1.3 meter long x 8 cm wide, with virus-free and BYDV-inoculated plants. Roots were washed at harvest and the root distribution was observed. Susceptible check Kyle had overall a slightly larger biomass of roots throughout the length of the tubes. All lines had a reduction of roots from BYDV infection. However, the interspecific line kept a better capacity of branching in the 50-69 cm deep area, which led to a maintenance of root mass despite the BYDV infection (Fig. 5). Some root rot, probably seed-transmitted, was visible on roots of 82PcDuros-476. We feel that more studies on root health are needed to better understand the observed performance of durum germplasm in BYDV inoculated conditions.

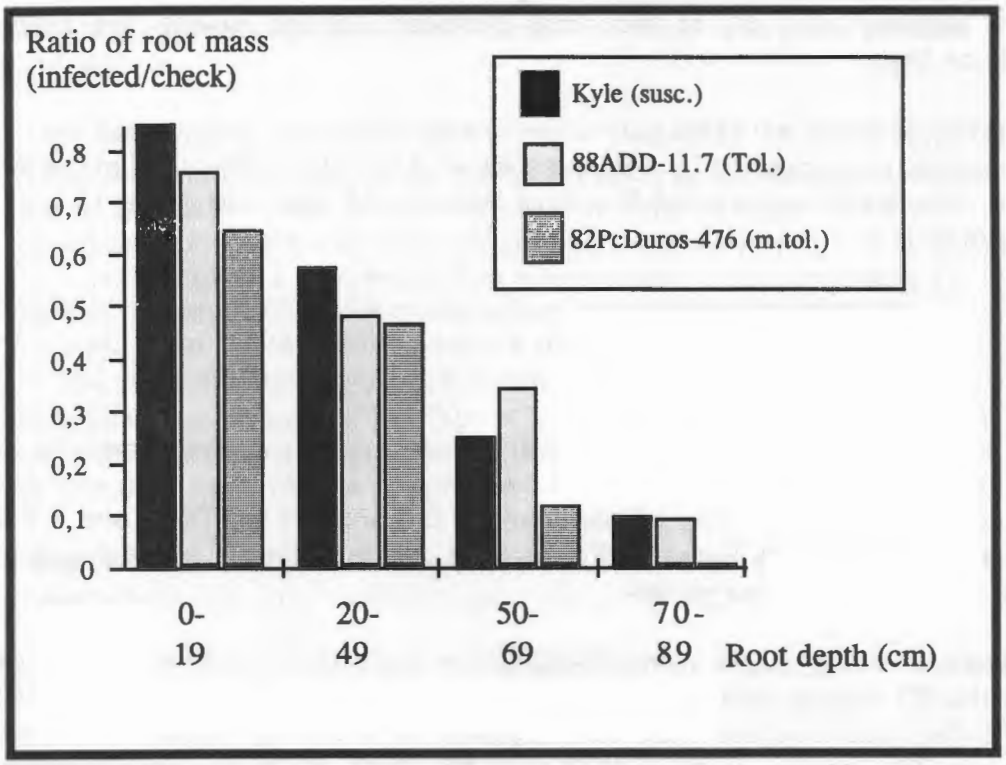

Fig. 5. Ratio of "BYDV infected / check" root biomass at various depth for susceptible durum Kyle and two BYDV tolerant durum lines.

Conclusion. Our current observations on BYDV, root rot and scab confirm that the cross 88 ADD-11 has yielded, after a long, tedious selection process involving artificial infection of segregating populations with BYDV, a very small percentage of exceptional disease-tolerant germplasm. This germplasm may be useful in breeding durum wheat with better disease resistance, which might help to change the area of adaptation of this species, and for example make it more adapted in Manitoba, where BYDV and scab are more common. The segregation of resistance and potential usefulness of the 88 ADD-11 lines should be evaluated in test crosses to cultivated durum lines.

Study and identification of markers of two genes for resistance to BYDV derived from *Thinopyrum intermedium*

C. Etienne, J. Jahier, D. Barloy and M. Trottet

INRA, Station d'Amélioration des Plantes, BP 29, 35653 Le Rheu, France

Two genes of partial resistance derived from *Thinopyrum intermedium* have been studied. The first one derived from an addition line of chromosome $7Ag^{i}$ in a Vilmorin 27 background extracted from a backcross progeny of a partial amphiploid (TAF46, 2n=56), created by Y. Cauderon. A translocation $7D\text{-}7Ag^{i}$ was produced by P. Banks (line TC14 in a Sunstar background). The second was found in the *Agrotricum* Zhong 5 from China, the resistance has been located on a homoeologous group 2 chromosome by P Banks (addition

line Z6). In the progeny of a F1 of the cross between Z6 and wheat, we selected ditelosomic addition lines.

Seedlings of the different genotypes were inoculated when the second leaf was half emerged. Three aphids contaminated by a PAV isolate of BYDV were deposited at the basis of each seedling. The aphids were killed five days later. ELISA test was made 15 and 30 days after inoculation.

Both sources of resistance slow down the multiplication of the virus in the plants, and have an effect on the proportion of plants which escape contamination. More than a third of TC14 plants, about 15 % of plants with Zhong 5 resistance and less than 1% of the susceptible plants escaped contamination. For the contaminated plants we observed a significantly lower multiplication of the virus (optical density) in the plants derived from Zhong 5 than in the other one. The mean optical densities 15 and 30 days after inoculation respectively were 0.2 and 0.5 for Zhong 5 derivatives, 0.6 and 0.9 for TC14 and 1.8 and 3.1 for the susceptible control Sunstar. These data suggest that the mechanism of resistance is different for these two genes

Molecular markers were found using RAPD. Five molecular markers of the gene translocated on the 7D chromosome have been found. The data obtained from a F2 of the cross TC14 × Rendez-vous suggest that the fragment of the $7Ag^i$ chromosome could recombine with the 7D. This has to be verified from the study of the F3. Four markers of the chromosome $2Ag^i$ were found: one on the arm without any resistance gene, one close to the centromere, and two on the arm carrying resistance, one of them being probably in a distal position.

The introgression of these two systems of resistance into an agronomic background adapted to France is under progress.

In 1996, we implanted in the field a test of tolerance with plants contaminated by a PAV vectotype of BYDV and plants not contaminated protected by insecticides. We observed that the yield loss of the lines carrying the tolerance gene *Bdv1*, from Frontana (Anza, Frontana and an isoline of Thatcher), was lower than the one of the resistant line TC14. But all these lines were spring type and poorly adapted to our conditions, so it is difficult to choose the best system before the transfer of the resistance and tolerance genes into adapted genotypes.

Resistance to BYDVs in Chinese interspecific crosses

M. Henry,[1] M. van Ginkel[1] and Qi Shiyu[2]
1. CIMMYT Int., Lisboa 27, Apdo Postal 6-641, Mexico, D.F., 06600 Mexico
2. Crop Breeding Institute, Heilongjiang Academy of Agricultural Sciences, Harbin, Heilongjiang, China

Two spring wheat lines, New LongMai 15 and New LongMai 19 (for both, n = 42), were derived from crosses between *Triticum intermedium* and commercial Chinese bread wheat

cultivars by Prof. Qi Shiyu and tested for their resistance to four BYDV serotypes at CIMMYT.

The isolates used were PAV-Mex, RPV-Mex, MAV-Mex and RMV-UK. Ten plants were inoculated with ten viruliferous aphids for each BYDV isolate; two plants were maintained free of aphids to serve as healthy checks. Bobwhite and TC14 were also inoculated and served as susceptible and resistant controls, respectively. TC14 is an Australian wheat line that also contains a *T. intermedium* introgression on chromosome 7D (Banks et al., 1994, *Genome* 38:395-405). ELISA was performed 15 and 30 days after inoculation.

Virus titers were low in the two Chinese lines and in TC14 for the four BYDV isolates. Virus titers were moderate to high in Bobwhite. Plants were assumed to be infected when the optical densities (OD) in ELISA were twice the average of the healthy checks. The results are summarized in Table 1.

Table 1: Virus titers (ELISA) measured in three presumably resistant and one susceptible bread wheat lines after artificial inoculation with BYDV.

	Mean OD at 15 days	Mean OD at 30 days	Number infected OD > 2*healthy
PAV			
NEW LONGMAI 15	0.087	0.083	7
NEW LONGMAI 19	0.110	0.096	8
BOBWHITE	0.594	0.544	10
TC14	0.106	0.096	7
RPV			
NEW LONGMAI 15	0.123	0.129	2
NEW LONGMAI 19	0.125	0.111	1
BOBWHITE	0.449	0.485	10
TC14	0.047	0.055	0
MAV			
NEW LONGMAI 15	0.097	0.065	0
NEW LONGMAI 19	0.110	0.057	0
BOBWHITE	0.659	0.448	10
TC14	0.126	0.089	0
RMV			
NEW LONGMAI 15	0.034	0.032	0
NEW LONGMAI 19	0.034	0.016	0
BOBWHITE	0.264	0.706	9
TC14	0.035	0.040	0

The results indicate that the two Chinese lines are resistant to virus multiplication of the four BYDV serotypes tested. These lines have been included in the crossing activities of the Bread Wheat Program at CIMMYT in order to incorporate alien derived BYDV resistance into elite bread wheats. The resulting progenies will be made available to our collaborators. Further studies will attempt to combine this resistance and the resistance based on TC14, with tolerance genes such as Bdv1 described by Singh et al (1993, Crop Science, 33: 231-234).

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Resistance to BYDV in barley

M. Henry and H. Vivar*
CIMMYT Int., Lisboa 27, Apdo Postal 6-641, 06600 Mexico D.F., Mexico
* CIMMYT-ICARDA, same address.

Barley lines showing good tolerance to one or more BYDVs were screened for resistance to virus multiplication. Sixty-six lines including the susceptible and resistant checks Atlas 57 (susceptible), Atlas 68 (resistant, tolerant) were inoculated. Six plants per line were artificially inoculated with 10 aphids of the following BYDV isolates: BYDV-PAV-Mex, BYDV-RPV-Mex, BYDV-MAV-Mex. One plant was kept free of aphids and used as the healthy control. Plants were maintained in the greenhouse for 15 days. One leaf per plant was then collected to be tested by ELISA. Plants were assumed to be infected when the optical density (OD) in ELISA was more than twice that of the healthy controls.

ELISA titers obtained ranged from 0.175 to 0.909 for PAV, 0.109 to 1.375 for RPV and 0.164 to 0.886 for MAV. Three lines out of the sixty-four tested had very low ELISA titers for both PAV and MAV and high titers for RPV. These lines were:
Tinctoria (ARUPO*2/3/PI2325/MAF102//COSSACK/4/ALELI)
Guayaba (ATAH92/3/MARIS CANON/LAUREL//ALELI)
Gramacote (CLN-B/80 5138//GLORIA-BAR/COPAL/3/VIRINGA/4/ALELI).
These lines also showed tolerance in the field for the same BYDVs.
Twelve lines had low titers after inoculation with BYDV-RPV (0.111-0.249) but only three of these showed tolerance for RPV under field conditions. These three lines were:
Dumari (MOLA/ALELI//MORA/3/MOLA/SHYRI//ARUPO*2/JET/4/MOLA/ALELI//MORA)
Faique (LBIRAN/UNA80//LIGNEE640/3/BERMEJO/5/CM67-B/CENTENO//CAM-B/3/ROW906.73/4/....)
Madre Selva (ROLAND-BAR/EH11//ESC.II.72.83.3E.7E.5E.1E/3/ARUPO*2/...)

Many of the lines tested showed reduced or no symptoms in the field, but had moderate to high ELISA titers, suggesting that tolerance was the most common mechanism.

The tested lines are high yielding barley lines from the ICARDA-CIMMYT barley program. Many of them also have multiple disease resistance. More work has been undertaken to characterize the type of BYDV resistance/tolerance present in ICARDA-CIMMYT barley program to provide suitable germsplasm for areas of South America where the virus causes severe losses to barley production.

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Trial on the resistance of cereals to viruses in the South of Ukraine

L. I. Omelchenko and L. T. Babayants

Phytopathology and Enthomology Department, Plant Breeding and Genetics Institute, Oridiopolskaya doraga 3 Odessa, Ukraine

Resistance of grain crops to virus diseases has been an integral part of the research activities at the laboratory of Phytopathology and Entomology of the Plant Breeding and Genetics Institute for the last 15 years. Because of the high cost of aphid maintenance and reliability of natural BYDV infection, screening for tolerance to BYDV in wheat and barley was done under natural infection conditions.

A new field screening procedure was tested during various cropping seasons since 1996. Each genotype was planted in six 2.5 m plots. Three plots / genotype were kept free of aphids by insecticide applications and served as the healthy check for each genotype. The experiment was kept free of natural aphid infestation and pathogen infection for the rest of the season by spraying insecticides and fungicides when needed. Symptom reading were taken after flowering, using a four digits scale; the first digit for the percentage of yellowish or reddish leaf area, the second for the percentage of dwarfing, the third for the percentage of reduction in tillering, and the fourth for the harvest index. All indexes were estimated by direct comparison with the healthy check of each genotype. Three hundred genotypes of winter wheat and forty five of barley were tested under natural infection conditions. Leaf samples from selected genotypes showing BYD - like symptoms were also tested by ELISA to confirm the presence of BYDV. None of the tested genotypes were found free of BYDV. Ten percent of the wheats tested had similar tolerance to BYDV. Among winter wheat varieties, Albatros odessky, Fantazia, Krasunya, Nadiya, Odesskaya 265 could be classified as tolerant to BYDV infection. The level of tolerance of the durum wheat varieties such as Aisberg odessky, Delfin, Delta, was also high. A crossing programme was started to study the inheritance of the BYDV tolerance in the selected material.

Strains of barley yellow dwarf virus in European Russia

K. Mozhaeva, T. Kastalieva and T. Vasilieva

Group of Virology, All Russian Research Institute of Phytopathology, Bolschie Vyazemy, Moscow Region 143050, Russia

Severe epiphytotics of BYD were reported on oat and barley in many regions of the Non-Chernozem zone and in a lesser extent in the Central-Chernozem region in Russia between 1988 and 1991. A cereal sowing area of several millions hectares was infected by BYD, causing important yield losses for agriculture.

Since 1992, we study BYDV in the "All Russian Research Institute of Phytopathology". Isolation of virus, its diagnosis, disease monitoring and strain identification are our fields of

investigation. In collaboration with M. Shemyakin and Yu Ovchinnicov from the Institute of Bio-organic Chemistry of the Russian Academy of Science, monoclonal antibodies and corresponding horseradish peroxidase conjugates were produced. The antigen used for mice immunization and clone selection was isolated from natural infected material (Erockina T., Kastalieva T. Zuchtungforchung, 1995, v. 1, N 2, pp. 71-74).

BYDV serotypes composition in infected samples were analyzed using these monoclonal antibodies and polyclonal antibodies from AGDIA Inc. The samples of cereals, grasses and weeds were collected in European Russia between 1992 and 1997.

Rhopalosiphum padi, *Sitobion avenae*, *Schizaphis graminum* and *Metopolophium dirhodum* known as vectors of RPV, PAV, MAV, SGV, and RMV were observed. Those BYDV strains were found in inspected areas.

PAV and RPV were the predominant strains in the Non-Chernozem zone. MAV and SGV strains were also found but were less frequent. Besides these four serotypes, RMV was observed in Povolzhskiy region. PAV, MAV and SGV were detected in the Central-Chernozem region.

In 1993-1997, an evaluation of BYDV incidence was conducted in cereals crops from Moskovskaya Oblast (Central region). In 1994, detail studies were done in oat and barley plots at the Research Institute of Agriculture in the Central region of the non-Cernozem zone. The incidence of infection was assessed on oat and barley using visual observations and sample analysis by ELISA. Incidence in barley was not high. It fluctuated amongst plots between 3,3% to 8,6% according to visual evaluation and between 2,7 to 7,9% according to ELISA analysis. RPV was predominant with approximately 70% of the infected barley samples, while PAV was found in 40% of the samples. Approximately one third of the samples were infected by both PAV and RPV. Higher levels of infection were recorded in oats. Visual inspection suggested that 34% of oat samples were infected, however, only 19% of the samples reacted positiveiy in ELISA. The predominant BYDV serotype was PAV (68%), followed by RPV (26%). 22% of the infected oats had a mixture of PAV and RPV [K. Mozhaeva et al., *Selekziya I Semenovodstvo*, 1:21-24 (1997)].

Table 1. BYDV occurrence in several Russian regions

Zone, region	Serotypes
Non-Chernozemic zone:	
North region	PAV, RPV*
North-West region	PAV, RPV
Central region	PAV, RPV, MAV, SGV
Volga-Viatka region	PAV, RPV*
Central-Chernozem region	PAV, MAV, SGV
Povolzhskiy region	PAV, RPV, MAV, SGV, RMV
Urals region	PAV*
North Caucasus region	RPV, RMV

* - Only RPV and PAV were detected.

In the main European Russia, aphid-vectors of BYDV are present and there is always sources for primary virus spread to small grain crops (winter crops, grasses and weeds), suggesting that BYDV could be a serious problem in this region of the world. Serotypes composition of BYDV and the degree of cereal disease are dependent upon the aphid species present and indirectly upon the weather conditions influencing the life cycle and the spread of the aphids.

BYDV incidence and epidemiology of wheat in Shaanxi Province of China

Y.F. Wu and N.S. Wei

Department of protection, Northwestern Agricultural University, Yangling, Shaanxi 712100, China

Wheat is an important grain crop in northern China. Yellow dwarf disease caused by barley yellow dwarf luteovirus (BYDV) is a very important wheat disease, causing severe yield losses in 1966, 1970, 1973, 1978 and 1980 in Shaanxi. Since 1980, this disease occurs every year and has extended quickly to neighboring wheat regions. Preliminary results of aphid transmission experiments and enzyme linked immunosorbent assays (ELISA) indicated that GPV is the most frequent serotype, but PAV as well as mixtures are also found frequently on winter wheat. GPV and DAV are the predominant BYDV types in Shaanxi. In natural conditions, infection of wheat seedlings by GPV and DAV depends on the presence of different aphids vector species. GPV is usually transmitted by *Schizaphis graminum* and *Rhopalosiphum padi*, and DAV by *Acyrthosiphum dirhodum* and *Macrosiphum avenae*. GPV and DAV are the Chinese BYDV serotypes that present no reaction with antiserum to PAV, MAV, SGV, RPV and RMV.

Many years of observation showed that climatic variables are important epidemiological factors. If air temperatures remain relatively low in June, become higher in October or warmer during autumn, then increase quickly in March of the following year, in conjunction with drought, barley yellow dwarf disease occurs heavily. Possible explanations to this relation between BYDV incidence and epidemiology, are the air temperature that correlates with fluctuations of aphid population (number and time of appearance) in early spring, when aphid vectors, in particular *S. graminum* transmit GPV from weeds to wheat seedlings. Population density of *S. graminum* in March may be suggested as an indicator to forecast BYDV incidence and epidemiology. Weeds and over-wintering infected hosts as a natural reservoirs, such as *Avena fatua*, *Sorghum halepense*, *Stipa brachychaeta* and *Cynodin dactylon* play an important role in BYDV epidemiology.

BYDV acquisition and inoculation are associated with the occurrence and duration of characteristic stylet activities during phloem penetration by aphids

G. Powell and J. Hardie

Department of Biology, Imperial College at Silwood Park, Ascot, SL5 7PY, UK

Phloem-restricted luteoviruses are entirely dependent upon aphid vectors for transmission; they are acquired when the insects ingest phloem sap, and inoculated when they inject saliva into plants. Stylet penetration activities by aphids can be studied by making the insect and plant part of a DC electrical circuit to generate sequences of characteristic waveforms called electrical penetration graphs (EPGs) [Tjallingii, *Ent. Exp. Appl.* 24:731-737 (1978)]. We have used this technique to investigate the aphid stylet activities which are associated with acquisition and inoculation of an MAV-like isolate of BYDV by the grain aphid, *Sitobion avenae,* and an RPV-like isolate by the bird cherry-oat aphid, *Rhopalosiphum padi.* Wingless adult aphids were allowed up to 6h electrically-recorded access to a BYDV-infected oat plant, and then transferred to a test seedling for a 2 day period to allow for vector latency. Insects were subsequently transferred to a second test plant for electrical recording of stylet activities during a range of inoculation access periods. This was followed by transfer to a third test plant, where the aphids remained for a 4 day period. All test plants were assayed for BYDV infection using ELISA 4-6 weeks following aphid access.

Periods of intracellular stylet tip location are registered as potential drops during electrical recording; these may be short (all cell types) or sustained (phloem sieve elements only). Two waveforms occur during extended phloem sieve element penetration by aphid stylets; firstly E1, which may later change to E2 (E1 + E2) e.g. [Prado & Tjallingii, *Ent. Exp. Appl.* 72:157-165 (1994)]. Most aphids (98%) which subsequently transmitted either virus to one or more test plants showed E1 + E2 during penetration of the infected plant. Separate analysis of E1 and E2 durations suggested that E2 is associated with virus acquisition, as insects which transmitted to test plants showed longer total E2 occurrence on the source plant than non-transmitting aphids. Most aphids (86%) which transmitted to the test plant during recorded inoculation access showed either E1 alone or E1 + E2. Inoculation tended to coincide with longer periods of E1. However, some insects which transmitted virus showed E1 for <5min, and 14% of inoculations were effected by aphids that showed no E1/E2, indicating that transient phloem punctures occur which are indistinguishable from penetration of other cell types using EPG recording.

The results suggest that electrically-recorded transmission characteristics of BYDV-MAV (BYDV subgroup 1) and RPV (subgroup 2) are very similar: acquisition of both viruses by aphids requires sustained periods of phloem contact, but inoculation can occur during very brief punctures of phloem tissue. These findings are in agreement with a previous study, where transmission of BYDV-PAV (subgroup 1) was investigated [Prado & Tjallingii, *Ent. Exp. Appl.* 72:157-165 (1994)].

Interactions between BYDV and a parasitoid in a shared aphid vector/host

Petra Christiansen-Weniger, Glen Powell and Jim Hardie

Department of Biology, Imperial College at Silwood Park, Ascot, SL5 7PY, UK

The pest status of cereal aphids may be reduced by parasitoid wasps whose larvae develop inside their insect hosts. These natural enemies are being increasingly looked to and implemented in modern aphid control strategies, but interactions with virus transmission have so far received little attention. We investigated interactions between an MAV-like isolate of BYDV and the aphid parasitoid *Aphidius ervi* (Hymenoptera: Aphidiidae) in the shared aphid vector/host *Sitobion avenae* with surprising results [Christiansen-Weniger et al., 86:205-213 1998)].

The impact of virus presence became immediately apparent when observing the mortality of aphids that had been parasitised by *A. ervi.* Significantly more insects died before formation of the aphid mummy (the parasitoid pupal cocoon inside the dead aphid) when both virus and parasitoid were present than when the parasitoid was present without virus. In addition, aphid dissection revealed that parasitoid development was significantly delayed in the presence of virus (on the 14th day after oviposition 95% of *A. ervi* developing in virus-free aphids had reached the pupal stage, whereas only 39% of those developing in hosts carrying virus were pupae). The virus moves through the haemolymph from the gut to the salivary glands and may therefore interact with the developing parasitoid, but the physiological mechanisms responsible for the increased aphid mortality are unknown. However, the results indicate a low suitability of viruliferous *S. avenae* as hosts for *A. ervi.* Studies were then made on oviposition success in virus-free and viruliferous aphids, to determine whether host suitability could be recognized by the ovipositing parasitoid. Indeed, female *A. ervi* appeared to discriminate, with significantly more eggs being deposited in virus-free aphids (56%) than in viruliferous aphids (39%). But does the presence of the parasitoid affect transmission of the virus?. A daily transfer of parasitised aphids to virus-free host plants after the initial virus acquisition access period showed that virus transmission was not reduced by the presence of the parasitoid developing inside the aphid, at least up to a point close to host death and mummy formation.

Thus, while the transmission of BYDV by the aphid vector remained unchanged by parasitisation, parasitoids may selectively avoid oviposition in viruliferous aphids as retarded development and/or premature death may result. Such observations indicate that the success of aphid parasitoids as control agents for BYDV in the field may be restricted.

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Name	Address	City&State/County/Department	Country	Tel; Fax; E-mail (Internet)
Javed Ahmad	Wheat Breeding & Genetics, Wheat Research Institute, AARI, Jhang Road	Faisalabad	PAKISTAN	Tel: +92 41 651 529; behzad@javed.fsd.brain.net.pk
Main research topics :	*Breeding for resistance & tolerance to BYDV in wheat, barley & triticale*			
Ligia Ayala	CIMMYT, Lisboa 27, Col. Juárez, Deleg. Cuauhtémoc, Apdo. Postal 6-641	06600 Mexico, D.F.	MEXICO	Tel: +52 5 726 90 91; Fax: +52 5 726 75 58/59; layala@cimmyt.mx
Phillip M. Banks	Queensland Wheat Research Institute, P.O. 2282	Toowoomba, QLD 4350	AUSTRALIA	Tel:+61 76 398 832; Fax:+61 76 398 800; banksp@dpi.qld.gov.au
Main research topics :	*Breeeding BYDV resistant spring wheats / BYDV surveys*			
Rosa Lia Barbieri	Departamento de Plantas de Lavoura, Faculdade de Agronomia - Universidade Federal do Rio Grande do Sul, Caixa Postal 776	95501-970 Porto Alegre	BRAZIL	Tel: +55 54 3166005; barbieri@ufpel.tche.br
Main research topics :	*Genetics of tolerance to BYDV in wheat*			
Muhammad Bashir	Pulses Improvement Programme, National Agricultural Research Centre, P.O. NIH	Islamabad 45500	PAKISTAN	Tel: +91 51 240468; Fax: +91 51 240909; pulses@sdnpk.undp.org
Main research topics :	*Studies on viral diseases of legume crops, Investigation on BYDV*			
Edmundo Beratto Medina	Instituto de Investigaciones Agropecuarias, CRI, Carillanca, Camino cajón-Vilcum km 10, Casilla 58-D	Temuco	CHILE	Tel: +56 45 215 706; Fax: +56 45 216 112; eberatto@carillanca.inia.cl
Lukas Bertschinger	Fruit-Growing Department, Swiss Federal Research Station for Fruit-Growing, Viticulture and Horticulture (FAW), P.O. Box 185	CH-8820 Wadenswil	SWITZERLAND	Tel:+41 1 783 6241(direct line); +41 1 783 6111; Fax: +41 1 780 6341; lukas.bertschinger@inbox.faw.admin.ch
Mohammed Boulif	Plant Pathology, National School of Agriculture, B.P. S/40	Meknes 5000	MOROCCO	Tel: +212 5 51 18 84; Fax: +212 5 51 16 55
Main research topics :	*Diseases of cereal crops*			
Vanderlei Caetano	EMBRAPA - CPACT, Caixa Postal 403	96001-970 Pelotas	BRAZIL	Tel: +55 532 758 437; Fax: +55 532 758 412
Main research topics :	*Tolerance to BYDV in wheat*			
Thomas Carroll	Plant Pathology, Montana State University, Leon Johnson	Bozeman, MT 59717	USA	Tel: +1 406 994 5154; Fax: +1 406 994 1848; upltc@msu.oscs.montana.edu
Oswaldo Chicaiza	INIAP, Estación Experimental Santa Catalina, Panamericana Sur Km. 18, Casilla 17-01-340	Quito	ECUADOR	Tel : +593-2-690 691; Fax: +593-2-690 991; chicaiza@sc.iniap.gov.ec
Frederick Michael Collins	New Farm Crops LTD, Market Stainton, Market Rasen	Lincoln, Lincolnshire LN8 5LJ	UK	Tel:+44 1507 343 348; Fax:+44 1507 343 526
Main research topics :	*Barley breeding*			
André I. Comeau	Canadian Agriculture Library, Research Centre, Agriculture and Agri-Food Canada, 2560 Hochelaga Blvd.	Sainte-Foy, Quebec G1V 2J3	CANADA	Tel: +1 418 657 7985 3212; Fax: +1 418 648 2402; comeaua@em.agr.ca
Main research topics :	*BYDV, Root health, Interspecific crosses, Haploidy*			
John Fletcher	Plant Protection, Crop & Food Research, P.B. 4704	Christchurch	NEW ZEALAND	Tel: +64 33 256400; Fax: 64 33 25 2074; fletcherj@crop.cri.nz
Allala Ghodbane	Plant Virology, Institut National de Recherche Agronomique, Rue Hedi Karray	2049 Ariana	TUNISIA	Tel: +216 1 230 239/1 230 024; Fax: +216 1 752 897
Main research topics :	*Plant resistance and tolerance to BYDV, Interactions BYDV - other diseases*			
Omar Guerrero Guerrero	Dept. of Plant Protection, Instituto Colombiano Agropecuario, Calle 19 A No. 42 A 45	Pasto	COLOMBIA	Tel. +57 927 233 812 / +57 927 221 162; Fax: +57 927 233 812 / +57 927 221 162
Main research topics :	*Plant Protection*			
Susan Halbert	Division of Plant Industry, Florida Department of Agriculture & Consumer Services, P.O. Box 147100	Gainesville, FL 32614-7100	USA	Tel: +1 352 372 3505; Fax: +1 352 334 0737; halbers@doacs.state.fl.us

Name	Address	City&State/County/Department	Country	Tel; Fax; E-mail (Internet)
Main research topics :	*Aphids, Taxonomy, Epidemiology, Field survey+B43*			
Richard Harrington	Entomology & Nematology, IACR-Rothamsted	Harpenden, Herts AL5 2JQ	UK	Tel: +44 1582 763133; Fax: +44 1582 760981; richard.harrington@bbsrc.ac.uk
Main research topics :	*Epidemiology, Vector, Modelling, Forecasting*			
Monique Henry	CIMMYT, Lisboa 27, Col. Juárez, Deleg. Cuauhtémoc, Apdo. Postal 6-641	06600 Mexico, D.F.	MEXICO	Tel: +52 5 726 90 91; Fax: +52 5 726 75 58/59; mhenry@cimmyt.mx
Louis Hesler	United States Department of Agriculture, Agricultural Research Service, 2923 Medary Ave.	Brookings, SD 57071	USA	lhesler@ngirl ars.usda.gov
Main research topics :	*Resistance to R. padi & BYDV*			
Winfried Huth	Plant Virology Dept., Federal Biological Research Center, Institut for Agriculture and Foresty, Biochemistry and Plant Virology, Messeweg 11/12	D-38104 Braunschweig	GERMANY	Tel: +49 531 2993 710; Fax: +49 531 2993 006
Istituto Fitovirologia Applicata	Area della Ricerca di Mirafiori - CNR, Strada delle Cacce 73	10135 Torino	ITALY	
Dragica Ivanovic	Plant Protection, Maize Research Institution, Slobodana Bajica 1, Zemun Polje, P.O. Box 89	11081 Belgrade	YUGOSLAVIA	Tel: +38 11 2356 704;Fax: +38 11 2356 707; djelovac@mrsys1.mr-net.co.yu
Main research topics :	*Maize virus diseases / Breeding*			
Makoto Kojima	Faculty of Agriculture, Niigata University, Ikarashi, Ninocho 8050	Niigata 950-21	JAPAN	Tel: +81 25 262 6600; Fax: +81 25 263 1659; kojima@agr.niigata-u.ac.jp
Main research topics :	*Genome analysis of Japanese barley yellow dwarf virus isolates*			
Mathias F. Kolding	Private 1910 SW 44	Pendleton, OR 97801-4221	USA	Tel: +1 541 276 0274
Main research topics :	*Breeding Wheat & Triticale*			
Alemu Lencho	Dept. of Plant Pathology, Plant Protection Research Center, P.O. Box 29	Ambo	ETHIOPIA	Tel: +251 1 360 094
Main research topics :	*Variability, frequency and distribution of BYDV Isolates on Wheat in Ethiopia*			
Abashamo Lencho Wako	Crop Protection, Pathology, Sinana Agricultural Research Center, P.O. Box 208, Robe-bale	Robe	ETHIOPIA	Tel. +251 06 610 271
Main research topics :	*Survey and identification of BYDV and aphid vectors*			
Sun Lianfa	Wheat Breeding Section, Crop Breeding Institute, Heilongjiang Academy of Agricultural Sciences, 368 Xuefu Road	Harbin 150086	CHINA	Tel: +86 451 666 8739; Fax: +86 451 666 3726
Main research topics :	*Breeding for BYDV resistance*			
Richard M. Lister	Dept. of Botany and Plant Pathology, Purdue University, 1155 Lily Hall of Life Sciences	West Lafayette, IN 47907	USA	Tel: +1 317 743 2197; Fax: +1 317 494 0363; lister@btny.purdue.edu
Simon McKirdy	Plant Pathology, Agriculture Western Australia, Locked Bag No.4, Bentley Delivery Centre	Perth, WA 6983	AUSTRALIA	Tel: +61 8 9368 3261; Fax: +61 8 9367 2625; smckirdy@infotech.agric.wa.gov.au
Main research topics :	*Control of BYDV spread, Screening for tolerance/resistance*			
Karine Moshaeva	Group of Virology, All Russian Research Institute of Phytopathology, Bolschie Vyazemy	143050 Moscow Region	RUSSIA	Fax: +7 096 334 0902
Main research topics :	*Purification of BYDVs, Diagnostic, Epidemiology*			
Paul M. Muthangya	Barley Research, Kenya Breweries LTD, P.O. Box 707	Nakuru	KENYA	Tel: +254 37 211 523; Fax: +254 37 212 998
Main research topics :	*BYDV Epidemiology and control, Resistance & tolerance to BYDV, Aphid vector monitoring*			

Name	*Address*	*City&State/County/Department*	*Country*	*Tel; Fax; E-mail (Internet)*
Larisa I. Omelchenko	Phytopathology and Entomology Department, Agricultural Sciences Plant Breeding and Genetics Institute, Oridiopolskaya doroga 3	270036 Odessa	UKRAINE	Tel: +380 482 69 45 12; Fax: +380 482 65 61 87 / 65 70 84
Main research topics :	*Resistance to viruses diseases of cereal crops*			
Ruggero Osler	Biologia e Difesa Piante, Universitá di Udine, Via delle Scienze, 208	33100 Udine	ITALY	Tel: +39 432 558 503; Fax: +39 432 558 501
Main research topics :	*BYDV in maize, rice and barley, BYD Epidemiology, BYDV and vectors*			
P. Peltonen-Sainio	Department of Plant Production, University of Helsinki, P.O. Box 27	FIN-00014, Helsinki	FINLAND	Tel: +358 708 5353; Fax: +358 708 5463
Glen Powell	Department of Biology, Imperial College at Silwood Park	Ascot, Berks SL5 7PY	UK	Tel: +44 1344 294242; Fax: +44 1344 294339; g.powell@ic.ac.uk
Dale Reeves	Plant Science Department, South Dakota State University, Box 2140-C	Brookings, SD 57007-2141	USA	Tel: +1 605 688 4756; Fax: +1 605 688 4452; kleinjan@mg.sdstate.edu
Main research topics :	*Oat cultivar development, Grain quality, Crown rust resistance*			
Robert H. Symons	Department of Plant Science, Waite Campus, University of Adelaide	Glen Osmond, SA 5064	AUSTRALIA	Tel: +61 8 8303 7423; Fax: +61 8 8303 7102
Main research topics :	*Molecular virology, Cloning Yd2, BYDV resistance gene*			
Maxime Trottet	INRA, Station d'Amélioration des Plantes, BP 29	35650 Le Rheu	FRANCE	Tel: +33 2 99285148; Fax: +33 2 99285120; mtrottet@rennes.inra.fr
Main research topics :	*Breeding for resistance and tolerance*			
Anne Wangai	Kenya Agricultural Research Institute, National Plant Breeding Research Centre	P.O. Box Njoro	KENYA	Tel: +254 037 61120; Fax: +254 037 61576; karinjr@arcc.or.ke
Yunfeng Wu	Department of Plant Protection, Northwestern Agricultural University	Yangling, Shaanxi 712100	CHINA	Tel: +86 0910 7093193 (H); Fax: +86 0910 7092559

International Maize and Wheat Improvement Center
Centro Internacional de Mejoramiento de Maíz y Trigo
Lisboa 27, Apartado Postal 6-641, 06600 México, D.F., México